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RESERVE FOOD-MATERIALS IN BUDS AND  
SURROUNDING PARTS.

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The purpose of this paper is to consider the structure and reserve food-contents of the buds and surrounding parts in some of our trees and shrubs, with occasional reference to nourishing substances as stored in other parts of perennial plants.

Particular attention will be paid to starch, because this is one of the most important constituents of the assimilated food of plants, is stored away in a granular form and admits, by means of its pronounced and characteristic reaction with iodine, of being easily detected and definitely located in the tissue bearing it.

Buds are the free extremities of branches, or, to place the same idea in a different form, they are incipient branches, whether located upon the free extremity or along the side of a stem. In the former case they are called terminal, and in the other lateral buds. As to their relation to growth they may be active, that is undergoing elongation or branch formation, or dormant, as illustrated by them in winter. It is with these last that this paper will have most to do, for it is in preparation for the inactive period that buds become most highly developed, and their tissues charged with the nutritive food-elements that are so much needed to push the plant forward during the unfolding in spring. Buds are again divided, as to their future development, into those that will produce



blossoms, called flower-buds, and those destined to grow into ordinary stems, and to bear the foliage of the plant. One set of buds become specialized, and are devoted to the particular function of reproduction, while their neighbors continue their growth and maintain the plant itself. The leaf-bud looks only to the future of the individual, while the flower-bud carries with it the idea of a new plant that in time is to flourish separately. In external appearance the buds of various trees and shrubs differ so greatly that they may well form a part of a full description of any plant. It is not our purpose, however, to go into any consideration of their sizes, shapes, colors and other peculiarities. Those buds that are herein treated have first of all a covering of scales accompanied by more or less of a lining of fur, and frequently in addition a varnish, all of which serve the important purpose of preventing access of water and the sudden changes of temperature that might otherwise bring death to these seats of vitality. The protective layers that a plant places over the tender substance of a growing point suggest the same provision for its own safety when hard times come as do the thick, impervious coats that are wrapped round the germ of a future plant as found ensconced in the seed. Within the bud-scales, which may be few or many, thick or thin, etc., there is the point of growth now in a quiescent condition. It is somewhat conical, possibly almost flat-topped, but from below it and upon all sides there arise a number of lateral outgrowths, in regular order, the youngest being nearest to the top, and these curve upward and overtop the growing point. These greatly disguised leaves are the first ones to unfold when the bud develops into a stem, while at the same time new ones are produced in close proximity to the advancing point of growth, thus continuing the bud as it pushes forward, leaving behind a young stem with its developing foliage. In case of the flower-bud, if it be one formed in autumn, as that of the apple, peach and similar fruits, there are the same bud-scales, but instead of a growing point that will develop into a branch, there may be one or several miniature flowers which, when the scales are thrown back, quickly open out into the fragrant blossoms.



It is a matter of common observation that buds, and particularly those producing blossoms, open with rapidity, burst, as it is termed, and there is no corresponding formation of substance out of which to grow at that period in the life of the plant. It follows, therefore, that there must be a reserve of formative material stored somewhere in the plant for the purpose of supplying the needed nourishment at this important and dependent annual period in the life of every tree and shrub. If we should consider seeds as to their minute structure, we should find that besides the plantlet there is frequently a large mass of lifeless substance stored in contact with the embryo, as in corn, and often enveloping it. This liquifies in germination and yields to the unfolding plantlet ample support until it can thrust its roots into the soil, lift its fresh leaves up to the air and sunshine and shift for itself. In many seeds, as the bean, the embryo is large and gorged with the food-material that the parent plant has provided for it.

There are many points of agreement between a seed and a winter bud. Both are constructed for a period of inactivity, and protected from untoward influences by thick layers of impervious substance. Both start into growth under the same quickening influences of vernal warmth and moisture, and in fact the buds of some plants, as those of the Indian shot-lily, regularly become detached, fall to the ground and are the starting-points of new plants. This is an important point in proof of the similarity of structure of seeds and buds. We should, therefore, naturally infer that at some place in every bud, or near by, there is a reservoir of those substances that are needed for its initial growth at least. A study of this point is now before us.\*

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\* The investigations, the results of which are herein presented, were begun during the winter of 1888-89, when, continuously for two months at the Iowa Experiment Station, the reserve food-substances in apple-twigs were studied in a score or more sorts, including a wide range of hardy and tender varieties. The results as then obtained appeared in Station Bulletin No. 4. During the past year, in spring, summer, autumn and winter, the work has been extended to include a large number of species of ligneous plants and the roots of many perennial herbs.



## STRUCTURE AND CONTENTS OF TERMINAL BUDS.

The terminal buds seem the most natural place to begin the consideration of the subject before us. In a general way, in the introductory notes, the structure of these buds, situated at the ends of shoots, has been pointed out, and it is for us now to pass more deeply into the subject, and with the aid of the microscope determine the minute anatomy of these points of conservation of vital energy. As before said, the leading component parts are the overlapping bud-scales provided with more or less hair and varnish, within which is the bud proper, that is, the incipient shoot, bearing its leaves as minute outgrowths which are arranged in a definite (acropetal) order; the large outer ones inclose the younger and smaller ones, the central point of all being the extremity of the stem. As in all newly formed vegetable tissue endowed with the subtle power of further growth, the cells that compose the central portion are small, thin-walled, and their contents consist largely of that colorless semi-fluid and extremely variable substance called protoplasm, the acknowledged vehicle of vital activities. Below this cone of delicate and almost homogeneous cellular tissue the stem is seen to take its origin with the differentiation into the cylinder of wood cells and vessels, having upon its inner side the comparatively unchanged pith, and, without, a thin layer of active cells that is to continue the growth in size of the branch by developing upon its inner surface new layers of wood, while outside is produced a protective substance familiarly known as bark. Therefore, by mentally combining transverse and longitudinal sections of the base of a terminal bud, the observer is able to determine the method of formation of the various elements that go to make up an ordinary twig. The young leaves that are a part of the dormant bud while still very small present but little more than the simple cellular condition common to the short tip of the branch that bears them. Among the outermost it is possible to distinguish the details of form and outline, together with the venation and other points of differentiation determined by a microscopic examination of newly unfolded leaves. Of course, the



distance to which this work of bud-growth extends, varies greatly within and outside of the species, and is not a matter to concern us farther at the time.

#### RESERVE FOOD-MATERIALS.

Plants which live from year to year do not use up all the nourishment prepared by the green parts, principally the leaves, during the season of active growth. They lay by a portion of this material to be employed in the vital processes at times when the plant cannot assimilate the crude substances which are obtained from the soil and the air. In short, perennial plants, during the growing season, store some of their elaborated substances in places where it becomes available for nutrition in the early spring, while the plant is putting forth its young twigs and leaves. This reserve material, as it is termed, may for convenience be divided into two groups: namely, those which are known as carbohydrates; so called because consisting of carbon and the elements of water, that is, of carbon, hydrogen and oxygen, united in definite proportions. The leading carbohydrate is starch, familiar to every one as the basis of many foods for animals and man, as found in potatoes, corn, and a long list of other vegetable products. Oil is another reserve form assumed by the carbohydrates, and abounds in many seeds and other parts of plants. Sugars, which by themselves make up a group of the carbohydrates, are often found associated with the other forms of reserve food-material above mentioned. There are other forms of carbohydrates, but they do not specially interest us in this paper.

The second division of reserve material, suitable for plant nutrition, is known as albuminoids, so named from a resemblance to the albumen or white of egg. Protein is another term given to the same group of substances, all members of which agree in having nitrogen in their composition—a substance which is absent in the carbohydrates. They are more complex and less stable compounds than the carbohydrates, and are stored usually as amorphous contents of cells. Sometimes, however, they assume the form of grains (aleurone), or crystal-like bodies (crystalloids), and in these condensed conditions may be met with in seed, like beans and peas, which are



usually rich in the albuminoids. The proteids are the basis of protoplasm, and protoplasm is the substance which is invariably present in every living cell. As protoplasm is the complex compound in which life always manifests itself, the importance of the albuminoids out of which protoplasm is made, becomes self-evident.

Doctor Vines, in his new work,\* in treating of the repose of the above compounds, says: "When once deposited the reserve materials undergo no change, or, at most, the proteids may slowly undergo some alteration, so long as the organ in which they are deposited remains in an inactive condition. An organ in this state is practically dead for the time being, all its metabolic processes being arrested. It is capable, moreover, of resisting injurious influences, such as extremes of temperature and desiccation, which would prove fatal to it, were it actively living. It is obviously in consequence of this property possessed by such organs during what we may term this state of suspended animation, that vegetation is maintained in regions in which the cold of winter is severe, and in arid tropical regions. The time of the possible duration of this state, without permanent loss of vitality, varies very widely." It is well known, for example, that some seeds retain vitality for a long time, especially starchy ones. With the quickening influences of warmth and moisture supplied by spring-time, the reserve materials undergo changes which convert them into substances that can readily travel to seats of vital activity when they are employed in growth.

*Starch.*—Returning to the carbohydrates, they will be taken up in the order of their importance. The test for the detection of starch is the very satisfactory one of iodine solution, which turns this substance blue, while it fails to produce the same color in other cell contents. By means of this reagent, starch is found quite generally at a short distance below the growing point, and usually in the form of compound granules, the component parts of which separate quite easily. These masses, or compound granules, vary greatly in size, and the smaller particles of starch do not exhibit this compound nature. Starch-bearing cells of well-matured twigs may con-

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\* "Physiology of Plants," 1886, p. 172.



tain hundreds of these granules, when they are completely filled with the nearly spherical masses.

The relative position of the starch varies greatly with different buds, especially in widely separated species of trees and shrubs, but in those that are well matured, upon the same plant, there is a fair degree of constancy.

At this point it is necessary to state that there are two classes of terminal buds, so called, among ligneous plants, with some gradations between them, not a little confusing. There are those which are characteristic of plants, with a well-defined habit of growth, that is, those that lengthen their shoots for a definite period, as in the horse-chestnut, for example, and then spend the balance of the growing season in forming a large terminal bud, and supplying it with nourishing food for the next year's development. On the other hand, there is a large group of trees and shrubs, and especially brambles, that grow on indefinitely, produce no plump, terminal bud for winter, usually kill back, and therefore must start the farther elongation of the twig from some lateral bud perhaps a foot or more back from the end of the stem. It is needless to say that the remarks to follow obtain only with the first class, and the consideration of the second will appear later, because less simple.

In order to make a careful, microscopic study of the cell elements of a terminal bud, it is necessary to secure several thin sections with a sharp razor, made in various directions through the bud. One of the most satisfactory is made lengthwise through the middle of the bud, and from this the position of the food-elements, after a little experience, can be determined with a fair degree of accuracy. This is said, not to discourage the making of horizontal sections, which are necessary for the demonstration of some important points, but to facilitate the work of those who may choose to extend the investigation herein outlined.

Figure 1 shows such a section through the terminal bud of an apple-twig, made in February, 1890, and therefore during a remarkably warm winter. The feature to be first observed is the dark column in the lower center (a) of the figure. This is the pith of the subadjacent stem, the cells of which are



starch-bearing ; this fact being indicated by the darkness in imitation of the color produced in this part of the section by a solution of iodine. At b and c are the scars of the last two leaves of the previous season, and from them pass inward and downward the vascular tissue that gives elasticity and strength to the twig. Other similar but smaller vascular threads are seen above, receding from the bases of the bud-scales, and uniting into a cylinder of woody tissue, still in a formative condition. The remaining part of the bud consists of small, many-sided cells packed together, a large number of them bearing complex, sphere-shaped crystals of oxalate of lime, while others are filled with semi-solid substances composing an impure form of protoplasm. The feature of most interest in this connection is the sharp line separating the starch-bearing cells of the pith from those in the younger pith just above, in the contents of which starch is entirely absent. There is a definite localization of the starch, as a rule, in or near all terminal buds that are fully matured. In those gathered before the processes of growth are completed, the starch may be scattered in all parts of the soft tissue, and especially in that portion lying between the zone of wood and the rind.

Besides the presence of starch, there is another marked difference between the pith indicated by the shading and the younger portion above, containing the albuminoids and crystals. The latter, it has been said above, is composed of thin-walled cells, and not materially different from those in all other parts of the bud. But the starch-bearing cells have their walls much thickened, and in such a manner that canals are left extending from the original cell wall to the free interior. These canals of adjoining cells meet at the juxtaposed walls, so that provision is thus made for the ready transfer of material from one part of the storage tissue to any other. In figure 2 is shown a portion of the tip of the starch-bearing pith in the terminal bud of a Duchess pear, and the adjoining thin-walled pith. At this line, which is usually somewhat convex, the tissue will often give way in making the sections, leaving the firm, thick-walled pith with a ragged edge. A few of the starch-bearing cells, highly magnified to show the details of their canaliculate structure, are shown in figure 3.



In making a longitudinal section of a terminal bud with a sharp knife, the operator quickly learns to detect the starch pith as soon as it is reached in the downward passage of the instrument. The bud proper is cut with ease, but there is a sensation known to grafters and others as that of "grittiness," which is due to the resistance to the knife caused by the many thick walls of the starch-bearing cells. This is one of the most important modifications of the ordinary cell wall, and consists of a thickening due to the intercalation into it of a substance called lignin, which chemically differs somewhat from cellulose, and adds materially to the hardness and durability of the tissue lignified. This power of resisting external influences is gained, however, at the loss of much elasticity. Lignified cells do not abound in protoplasm, but water passes freely through their walls. All permanent, inactive tissue may become lignified, and when this process is freely carried out it yields the durable heart-wood so familiar in many kinds of timber. The subject is of interest because it helps to explain the matter of "grit," or "grittiness," so frequently spoken of by those who cut the twigs of various sorts of fruit-trees. The inflexibility of tips of certain plants is due in most parts to the large amount of lignin their pith contains.

The free end of a mature twig consists of a cone of minute, thin-walled cells, upon the outside of which the small, imperfect leaves, as bud-scales, arise in regular order, and, overlapping each other, enclose the tender growing point of the twig. From the scales, bundles of fibers and vessels descend and form a thin ring of wood around the pith just below the growing point. At this portion of the twig the pith makes up the greater part of its substance. In short, the bud, the stem, and for a quarter to a half inch below it, are composed of soft tissue, easily crushed with the thumb and finger. But below this the large central pith is particularly rigid, due to the unusual thickening that has taken place in the walls of the cells. If the reader will bear in mind that the wood zone is very thin at the upper part of the twig, and that the bast is almost wanting, it will be evident that the pith alone must give the rigidity found near the extremity of matured twigs. From the shape of the nearly spherical cells it follows that



there can be no very great toughness. Contrariwise, brittleness characterizes the upper portion of a well-matured twig, and for two or three inches below the terminal bud it will often snap, almost like a pipe-stem, when sufficient side pressure is applied.

*Sugars.*—Tests for other carbohydrates were made, principally the various sorts of sugars.\* Grape sugar was generally present in variable quantities in all terminal buds. Cane sugar and dextrine were likewise present in most cases, but in small quantities. Grape sugar was most evident near the growing points, and cane sugar, if it appeared at all, was in the fine, unlignified tissue at the base of the bud. In view of the fact that starch is readily changed into sugar within the plant, under conditions similar at least to those surrounding the twigs when gathered for testing, it follows that sugar, which in this connection may be considered as another term for soluble starch, would be expected, and in variable quantities. Twigs gathered directly from trees also showed these sugars in small amounts.

*Albuminoids or Proteids.*—This group of complex and very variable substances does not readily admit of separation into individual sorts, and is therefore treated as a whole. Proteids are present in all living parts of ligneous plants, and are most abundant near the points of greatest vital activity. In the apple, for example, there is little or no albumen in the pith at the base of an old twig, very little in the wood zone, an abundance in the cambium layer, and the largest percentage in the buds. In these last places, when the buds are mature, the albuminoids make up a large part of the cell contents, to the exclusion of the starch. All the varieties studied exhibited the albuminoids as brick-red particles when treated with the Millon test, a nitrate of silver compound, described elsewhere. On account of the conspicuous color the presence of the protoplasmic compounds is easily demonstrated. When the tip of a twig was still soft, spongy and without rigidity from a failure of the pith to lignify, there was only a feeble response. Protoplasmic matter was present, but so scattered

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\* The methods observed in the micro-phytochemical study of these carbohydrates will receive separate treatment at the close of the paper.



over a large area of cellular tissue that no distinct line or point of coloration developed, as is true of buds which have become ripe.

The albuminoids, therefore, like the starch, from their behavior in green and ripe twigs, become a test of maturity. These complex compounds are the ones out of which protoplasm forms, and afterward renews its strength. They are the most important reserve materials in the sense that they stand, in composition, nearest to that of protoplasm, which is the acknowledged vehicle of vital activities. It is not unnatural that these compounds, in a resting twig, should be stored in the buds where they are to be needed the coming spring. Starch may, therefore, be excluded from a well-equipped bud, because other substances of greater consequence, and accumulated in smaller quantities, are conserved at the points of growth. In other words, there is a manifest tendency for the concentration of reserve materials at points where they are to be used, and in the order named; first the albuminoids, and next the carbohydrates—first the basis of protoplasm, and next the substance protoplasm must use in the building up of new tissue, and in other vital processes. Following upon this is the conclusion resulting from the investigations, namely: that, other things remaining the same, the best-conditioned twig is the one having a sufficient amount of reserve material within easy reach, and in the best condition for the use of the plant. A well-preserved bud is therefore plump with reserve substance in a comparatively solid condition, being firm but not woody; is well protected from the injurious effects of very sudden extreme changes of temperature, and has close at hand a sufficient amount of starch, or other carbohydrates, for its most advantageous development. Whether one form of bud, as to its length, breadth and thickness, color or number, and hairiness of scales, may be better than another under any special condition, is a question more likely to be determined by field trial than by laboratory tests. Plants, seemingly equally hardy, may have the vital points very differently constructed. No matter along what line the problem has been worked out by the species, the chief point is to prepare for hard times, and having once made the prepa-



ration remain quiescent until the coast is clear for another year of growth. The inherent tendency of one introduced variety of plant to start into growth before another may throw it into the class called tender, while another with the same structure, but differently disposed, will prove hardy. The test for sugar in the late winter may aid in deciding that a difference in this tendency exists, for starch, the chief form in which the carbohydrates are stored up in many kinds of twigs, is changed into sugar before it becomes available for plant nutrition.

*Crystals.*—A few words concerning these structures, which were constantly met with in all buds examined, will be sufficient for this portion of the subject. Vegetable physiologists are agreed that true crystals (not including crystalloids) in the tissue of plants are a form of refuse, or left-over matter, resulting from the processes of growth, and put up in a consolidated form, to get them as much out of the way as possible. Some one has compared them to the pieces of brick, mortar and other material thrown into boxes and barrels during the construction or repair of a building. Crystals were rarely found in the old pith, and have not been seen in the wood, but are very abundant just below the growing tips of all buds in that cylinder of tissue connecting the free extremity of the bud with the starch-bearing cells—a half-inch or so back of the tip. The loose green bark of all parts of the twigs also abounds in these bodies, and they are especially numerous in the cellular tissue that lies between the leaf scar and the bud above it. In this locality, a second form is often met with, which is smaller and rectangular in shape, while the prevailing sort is an irregularly spherical aggregation of sharp-angled bodies, which have taken the name of sphæro-crystals. These crystals are composed of oxalate of lime (calcium oxalate) and may be dissolved by mineral acids.

From the composition of these bodies, their universal prevalence in about equal numbers, first in proximity to tissues which are the centers of rapid vital processes, and secondly, in out-of-the-way places, and for various other reasons, it is safe to conclude that crystals are no safe criterion by which to judge of the relative resisting powers of plants to



the untoward influences that may surround them. One would as soon think of deciding upon the stability of a house or bridge by the number of chips that the builders have made.

*Tannin*.—Like crystals in vegetable tissues, tannin is considered as refuse matter, and not, with perhaps certain exceptions, further active in the vital processes of the plant. Tannin, as recognized by salts of iron, is abundant in twigs, and in transverse sections is found most common in the rind, and the inner bark close upon the cambium, and between it and the zone of bast. There is a great difference in the amounts of tannin between the several species. The buds contain more than the other portions, and in immature terminal buds the tannin is quite generally diffused throughout all parts of the soft tissue.

#### COMPARISON OF VARIOUS SPECIES.

The reader is now prepared to make a comparative study of the food-materials in the terminal buds of some of our leading kinds of trees and shrubs, confining his attention for the present to those with well-developed buds at the extremities of the branches. In the horse-chestnut we have a typical illustration of buds with large and therefore conspicuous parts. A longitudinal section is made with great ease, and there is noticeable absence of the grit found in many twigs. The pith is very large, and at an inch or so below the bud it is dry and filled with air. The starch deposit occupies a thimble or nipple-shaped portion of the moist pith, above which is the growing point with its proteid compounds. Instead of growing for a long distance at the least expense and strengthening the upper portion by a deposit of lignin in the pith, the wood zone continues of considerable thickness, and the buds rest in this ring as a crayon is held by its handle or clamp. Figure 4 illustrates these points better than can be described in words. The small amount of starch present may be due to the warm winter, for the varnish was much softened at the time of gathering (February 20), and the moist (almost wet) pith was filled with a mixture of the soluble carbohydrates and the albuminoids ready for the initial growth.

In remarkable contrast with the last are the clusters of



buds at the ends of twigs, as in the cherry, oak, etc. The tip of the twig is enlarged; the wood zone is thin, but there is a corresponding lignification of the great mass of pith, thus giving stability to the end of the shoot and furnishing a reservoir for a large supply of starch from which the several buds can draw liberally when they need this nourishment.

The maples offer a case of buds being in pairs, and therefore there are three very near to the extremity of the twig—the terminal or central one and two lateral buds. In the hard maple (*Acer saccharum*, Marsh.) the disposition of the starch suggests a thimble or a finger of a glove, it forming a narrow layer, in the pith that lies next to the thin zone of wood. In the soft maple (*Acer saccharinum*) the thimble has no tip, but is like a “glove finger” that has been cut off near the end. This may arise from the fact that the lateral buds are not so close to the terminal one.

The ashes are like the oaks in having a large amount of starch near the terminal buds. On the other hand, the catalpa resembles the soft maple in having a “thimble.”

In the locust we have an illustration of a poorly formed terminal bud, and the starch distribution is accordingly peculiar. The treatment of such cases comes more properly under the head of lateral buds, but they form a transition between the terminal and lateral. The starch of the upper two inches of the twig is distributed quite equally between the two buds either of which might receive it all. In the basswood there is much the same condition of things, and in the hackberry also.

Among shrubs the lilac is a striking example of copious starch-storing at the tips of the branches, but in this case it should be remembered that two large buds are located terminally and two shoots must be provided for instead of one. Figure 5 shows the quite constant condition of the shrub in winter, and as this plant has been studied throughout the year it will be treated of elsewhere.

The garden cherries have terminal clusters of buds as in the oak, only larger, but the amount of starch is small. The same is true of the wild cherries. However, a large amount of gum or mucilage is present, and this doubtless serves the same purpose as starch in the economy of the plant.



Plums have much more tapering twigs, and end in minute terminal buds, but with considerable starch located in a cone of lignified pith, as has been mentioned for the apple and pear. In the peach the terminal bud is large, but the starch is much less abundant than in the pear and apple; it is midway between the pear and the cherry in respect to its starch-bearing, and perhaps holds the same position as to the amount of mucilage or gum.

There are many species of trees that do not usually present any starch in the twigs of the last year's growth, among which are the various kinds of willows and poplars, but the older wood of the main branches, as a rule, contains more or less of this food substance. Among shrubs the elder and currant and gooseberry have but a small amount of starch in the vicinity of the terminal buds. In short, there seems to be no evident rule to guide one in the matter. Starch may be in abundance in one species when least expected and almost absent from another.

*Other Carbohydrates.*—An explanation of the absence of starch in many twigs is not far to seek. It may be due first to the substitution for it of oil, as in many seeds as well as buds and twigs. The pine family is a striking illustration of this, in the buds and branches of the members of which a pitch abounds, largely to the exclusion of starch. Sugars of the various sorts in like manner may take the place of starch in furnishing the carbohydrates needed for the growth of plants. In the second place, starch may have been deposited, but afterward changed into those closely related chemical compounds that, being soluble in cell sap, are ready for transportation and work. Rarely does a mature bud not respond to the Trommer and Fehling tests, but those without starch in their vicinity indicate the largest amounts of the soluble starches, as they are sometimes called. In this connection it may be said that the wood of the common currant bush was thoroughly examined because of a failure to find starch in the young twigs. When the roots, however, were tested, it was found that they abounded in starch. Albuminoids and sugars were present in the ends of the twigs, while the storehouse of starch was below ground.



## LATERAL BUDS.

In general structure the lateral buds closely resemble the terminal ones, but are much smaller, and have a somewhat different attachment to the twig. The soft cone of small cells rests upon an abbreviated stem from which the outer scales arise. Where the bud is connected with the twig there is an interlacing ring of fibers and vessels, and within this is a central pith, which is modified as spoken of while treating of the terminal bud; that is, the cells are very thick-walled, having undergone the process known as lignification, a characteristic of the durable parts of most ligneous plants. As the lateral buds are often close to the twig upon one side there is a consequent lack of symmetry. The exposed surface naturally has a greater development of the protective layers, the bud-scales being both thicker and more numerous upon the outside than next to the stem. There is also more of the red coloring matter in the exposed parts. Over all parts there may be a coating of soft down consisting of colorless hairs which grow from the epidermal or outer layer of cells of the bud-scales and the body of the twig. As the twig matures this hairiness is quite easily removed, so that its presence or absence in mid-winter is largely accidental.

*Flower-Buds.*—As a general thing, the conditions that surround a flower-bud are necessarily much the same as those of the leaf-bud. Both are incipient branches, and while one is destined to elongate into a twig, bearing ordinary foliage, the other remains comparatively short and produces disguised leaves for the purpose of reproduction. It is true, however, that while the leaf-bud soon becomes self-supporting, and in fact finally makes the starch for the next season's use, the fruit-bud is a source of loss to the plant, and therefore needs to be provided with nourishing food-materials. On this account it is natural to expect that the fruit-buds should be specially favored in the location of starch and albuminoids.

Flower-buds usually are not strictly terminal, that is, at the extremities of main twigs. However, there are exceptions, and the treatment of flower-buds does not fully fall under either of the two groups of buds already considered.



Thus in the peach the flower-buds are not at the ends of the shoots, but instead are here and there along the twig in the position of, and easily distinguished from, the lateral leaf-buds by their larger size, extreme hairiness, etc. Not infrequently there are three buds at a node and raised upon an abbreviated spur, but in such cases the central one is a leaf-bud, and the two lateral ones each contain a flower. The apple, in somewhat the same manner, has lateral spurs, at the end of which is a bud containing three or more immature blossoms apparently terminating the branch, but in fact only overreaching a dwarfed leaf-bud situated somewhere among them.

In the horse-chestnut is an illustration of the center of the large terminal bud being occupied by a flower-cluster which when unfolded is a foot or more in length, and bearing possibly a hundred blossoms. This flower-cluster, at first terminating the stem, soon takes a lateral position by the development of a leaf-bud that continues the twig.

A large number of flower-buds have been examined during the past winter, both to determine the disposition of the reserve substance and to make notes of the influences of the warm weather. As far as the storage of starch, sugars and albuminoids is concerned, it can be briefly stated that there is no material difference between the fruit and the leaf buds, except that the latter are usually larger, lack the light green interior, characteristic of the young parts of leaf-buds, and contain within and near them a greater supply of the albuminoids, backed up by a lignified starch-bearing tissue. Not being strictly terminal, the fruit-buds are well located for the immediate storage of starch in the wood and pith of adjoining parts.

In the fruit-buds of the peach there is an interesting case of the special localization of starch that has not been found in those of apple, plum, cherry, quince, or in fact any other blossom-buds subject to examination. Without entering into a description of the peach blossom, it may be said that there is a single pistil centrally located, about which the other parts are disposed—the calyx as a cup bearing the stamens upon its inner surface. In figure 6 a peach-bud is seen in longitudinal section, with its upper part of fuzzy bud-scales removed, exposing in the center of all the pistil a, and next the stamens b.



All of the base of the bud *c* is charged with albuminoids and carbohydrates that do not respond to the starch test. When the wood, *d*, of the twig is reached, it is found filled with starch, as indicated by the shading. Near the base of the pistil, or miniature peach, is a triangle of starch-bearing tissue. Occasionally, starch is also found in the pistil and the stamen-filaments. These last need to undergo rapid growth at time of blossoming. Whether this starch was stored in these parts, far away from the great reservoir of this material, during the growing season, or has been laid down afterward, is a question that is not answered. It is, however, a well-known fact, that starch can be deposited, then dissolved, and re-deposited in another place repeatedly within a short time when such a change of the carbohydrates is of advantage to the plant. A study of the formation of the callus, and of the knitting tissues of grafts, is interesting in this respect. It is, however, true, that when the flowers develop the reservoir at the base of the peach pistil becomes emptied of starch, and the tissue, not being thick-walled like ordinary starch-bearing cells, is similar to that around it.

### TRANSVERSE AND LONGITUDINAL SECTIONS OF TWIGS.

An ordinary twig of the last season's growth consists of the following parts: (1) A central cylinder of pith which runs the whole length, ending at the tip in the terminal bud. This pith consists of many-sided cells, which are about as long as broad, and packed so closely together as to leave but few cavities, called intercellular spaces, between them. The cells nearest the center are usually largest, and the outermost smallest, and have the thickest walls. (2) Next outside of the pith is a ring or zone of wood, which is very thin at the upper end of the twig, but gradually becomes thicker as the basal end is approached. This wood consists of long ducts or vessels running longitudinally in the twig, and surrounded by slender, thick-walled flexible wood cells. The wood is the most substantial part of a twig, and makes up the greater part of any tree. In the wood ring of the twig are thin plates of pith-like cells, which reach from the pith to a thin



belt of cells, capable of growth, situated just outside the wood, and called the cambium layer. These plates of thin-walled cells, known as the medullary rays, are here especially mentioned, because they play an important part in the storage of the reserve food-substance formed by the plant during the growing season, and stored away to be employed during the initial growth in early spring. Besides the vessels, wood cells and medullary rays in the wood ring, there are also long, rectangular starch-bearing cells, in many respects not unlike the pith, extending lengthwise of the wood, and at right angles to the general direction of the medullary rays. These may be styled the wood-pith cells. (3) The cambium layer, above mentioned, is made up of small, thin-walled cells, and constitutes the soft layer at which the wood and bark may be separated, especially in spring. The radiating plates of pith cells which reach from the pith to the cambium layer, and above designated as the medullary rays, project beyond the cambium, and broaden out in the loose, cellular part of the bark that lies between the somewhat interrupted ring of bast and the cambium. (4) This bast is the tough fibrous part of the bark, and consists of small bundles of thick-walled cells which run lengthwise of the twig, and are usually midway between the rind and the cambium. (5) Upon the exterior of all is a double layer of thick-walled cells. The outer may be styled the cuticle, and beneath this is a much thicker layer of firm tissue, with the contents of many of the cells colored green. The outer and thinner layer bears much of the coloring matter which gives mature twigs their characteristic reddish, brown or other color. Between the rind and the ring of bast is a belt of loose, cellular tissue abounding in cavities between the cells known as intercellular spaces. This is the loosest tissue in the twig, and may be called the pith of the bark. It, however, differs from the true pith in the center of the stem in having the cell contents colored green. It is often and appropriately called the green bark, and is the part exposed when the thumb-nail removes the rind of a twig but does not pass deep enough to reach the wood layer.

Therefore, to recapitulate, the twig in cross-section consists of the following parts, beginning at the outside: A double



rind or protective covering of thick-walled cells more or less impervious to water, and bearing the matter which gives the twig its color. Within this is a broad, loose belt of green cells and large intercellular spaces which reach to the tough fibrous band of bast. Succeeding the bast, passing inward, is another loose belt of cellular tissue narrower than the one outside the bast into which the medullary rays project as they pass through the cambium layer. Next, within this cambium, is the wood with its medullary rays which reach to the central cylinder of pith. The outer portion of the pith, the medullary rays, and the pith wood-cells at right angles to the latter, are the three portions of the internodial part of a twig that are starch-bearing. The above description considers only sections that might be made through the twig at any point between the buds. At the nodes or points where lateral buds are developed there are certain modifications of the structure which have been considered.

**JUNCTURES.**—At the point of union or junction of two years' growth of a twig, as of that found during 1889 and that of 1890, there are but few structural features not included in the description of the twig already given. The point is easily determined from the outside, for the scars of the several scales of the terminal bud of the previous year remain to mark the place, which is somewhat larger in cross-section than the twig, an inch or so above and below it. It also is a starting-point from which the buds become more distant, proceeding either up or down the twig. If these points are not sufficient, the brighter, fresher red of the more recent growth will be quite sure to distinguish the place of union. With a cross-section of the twig under the microscope, the age in years is quickly determined. The first wood formed on a yearling twig in spring is more porous than that last produced the previous autumn. It is on this account that the wood of an old branch is arranged in evident rings, and if there has been no interruption in the growth of the plant during any summer there will be a ring for each year, the thickness and porosity of which will vary with the season's favorableness for growth. Microscopic inspection of the juncture does not reveal any very marked modification of structure. The cylinder of pith is



larger here, often twice the normal diameter of that just above or below ; its cells are smaller and thicker-walled and inclined to be elongated and arranged in longitudinal rows, as if to add rigidity to this portion of the twig during the early spring growth, and give general stability at a point where elements of weakness naturally obtain from the union of the wood of two different years. Below the juncture the pith contains a cone or thimble, the outer cells of which are usually tinged with brown as if dead, and in shape and position it accords with the exterior of the lignified pith which has been mentioned as beginning a short distance below the base of the terminal bud. With certain chemical reagents and coloring materials this thimble or cone of pith, surrounded by structurally almost identical tissue, behaves the same as that at the top of a well-matured twig, and easily met with by cutting down through the terminal bud lengthwise with a razor or sharp knife. It is evident, therefore, that as the terminal bud undergoes development in the spring, the soft thin-walled cells at its base become lignified, and while they are finally apparently the same in structure as the cells below them which were lignified the autumn before, the line of separation is not obliterated, but instead may be usually observed, without the aid of a magnifier, by making a longitudinal section through the juncture.

The point that most interests us in this connection is the important part which the juncture plays in the storage of reserve material, and especially starch. Having become adapted for this service while situated in the vicinity of the terminal bud, the lignified pith cells continue for several years in the same capacity, and probably at no time become entirely free from these substances. Trees and all starch-bearing shrubs exhibit this fact, but some much better than others. For example, the pear is shown in figure 7 as a type of the large class of trees. Figure 8 is of the honey locust, which is without a well-formed terminal bud, and the branch is each year continued by a lateral bud. Twigs with opposite buds, which, of course, from what has been seen of the relation between buds and starch storage, have the amount of food-reserve doubled at any one transverse plane of the node, are



illustrated in the ash, figure 9. In the lilac, as before mentioned, there are practically two terminal buds, and this leads to another form of juncture shown in figure 10.

Some extremely slow-growing horse-chestnuts and ailanthus trees, found in a rocky situation near New Brunswick, N. J., present striking illustrations of the point in hand, because what is usually found in a twig of several feet in length is condensed into as many inches. The ailanthus, figure 11, illustrates the method of killing back of the tip each year, and the renewal from a lateral bud. In fact, everything has gone so irregularly that the storage of starch at the junctures is far from uniform. In the *Æsculus*, figure 12 a, the case is very different, and the progress, although small, is quite uniform. Looking at this twig from the outside, there is one almost continuous display of bud-scale scars, so that the age of the branch could be determined only with much difficulty. On the other hand, by splitting it through the middle, the empty, thin-walled, colorless sections of pith are quickly seen as alternating with those of a brownish tinge. But the greatest difference is manifest when half of such a twig is laid for a few moments in a dish containing iodine, and afterward washed in alcohol. It is then that the blue sections of pith are seen to correspond to the basal part of each successive terminal bud, and the empty pith separates them. The starch-bearing cells in the *Æsculus* are not as much lignified as in most twigs, and for this reason, when they lose their normal amount of moisture by exposure, will shrink, leaving depressions in the pith channel at these points, while the empty, colorless pith between them retains its plump form indefinitely. An examination of a similar *Æsculus* stem, made July 30th, showed that only a small fraction of the starch still remained, and with almost none in the bud itself. At b, in the same figure, the parts of a rapidly grown, long horse-chestnut twig are shown in contrast with the slowly developed branch at a.

#### SPINES AS RESERVOIRS OF FOOD.

A study of the winter contents of the thorns of honey locust, hawthorns, wild crab-apple and the Japan quince, etc.,



leads to the conclusion that these sharp means of defense serve another purpose, perhaps only secondary, besides that of protection. Upon examination for starch, it was found that there was an unusually large per cent. in the thorns. A thorn of a honey locust, for example, when examined in midwinter is found to consist of a horny exterior, within which is a dry, somewhat powdery, snuff-brown mass, containing no starch. Near the base of the thorn at that place where it is sometimes flexible in its attachment to the stem, when growing, for example, upon the bole of the tree, the central pith is firm throughout, of a greenish color when freshly cut, and packed with starch. In figure 13, at a, is shown a longitudinal section, through such a thorn. It is also seen by the same figure that the starch diminishes rapidly in amount, passing upward from the thorn, while below it remains of about equal amount until the lower of the series of buds below each spine is passed.

The position of thorns being near the bud favors the idea of their serving as places for the storage of starch, for when thus laid away for the winter it is close at hand for use when the growing days of spring arrive. The relative amount at the base of the spine, and below at the point bearing the buds, is shown in the cross-section b, and not only is there more within the stem at the juncture of the spine, but the base of the latter is more thoroughly gorged than any other part of the tree. At c is a section shown midway of the internode.

In the hawthorns the structure of the base of the thorn is somewhat different. Omitting the discussion of the minute structure of the spine it will be sufficient to call attention here to figure 14, in which the starch, the darkened portion, is seen to occupy the pith of the twig, making a V-shaped structure at the base of the spine. In addition to this, there is a somewhat triangular starch-bearing portion above and below the pith of the base of the spine. In some specimens the starch extends for nearly a half-inch into the spine, but as a slender point.

In the wild crab (*Pyrus coronaria*, L.), the spine is more like an ordinary branch, because usually bearing buds upon its



exterior, and has the starch-bearing pith extending for a half of the way, from the base to the sharp, hard tip. In the main twig there is a manifest accumulation of starch at the base of the spine, as is shown in figure 15. Figure 16 shows a fruit-spur, the center of which is filled with starch for the purpose of providing abundant nourishment for the development of the cluster of flowers.

Many other examples might be cited, but that of the Japan quince (*Cydonia Japonica*, Pers.) will suffice. The spines of this favorite hedge plant were examined in February, while some of the blossoms had already expanded, due to the very warm winter of '89-'90. The position and relative size of the parts are shown in figure 17. It will be seen that the flower-stem is short, and the spine joins it at the base. In figure 18 the condition of the starch storage becomes clear. There is no starch in the flower-spur, but an abundance in the interior of the spine, to be withdrawn to feed the flower and the leaves that afterward come upon the stem.

It is very likely that the spines are primarily for the warding off of enemies, but if we can look upon the protective organs as serving another purpose it adds further dignity to the police department, so to speak, of the plant.

#### METHODS AND REAGENTS.

STARCH.—For the detection of starch, as before stated, the solution used was iodine made as follows: three grammes of crystallized potassium iodide were dissolved in sixty c. c. of distilled water, to which was added afterward one gramme of metallic iodine. This solution was diluted as the occasion required. The microscopic inspection for starch was with thin sections either treated at once with the iodine solution or after they had received potassic hydrate to remove the protein compounds that might otherwise obscure the reaction. When the presence of starch was established its abundance and general distribution was determined by splitting the bud or twig, or both, and immersing the parts for a few minutes in a long porcelain trough, when the degree of darkening of the various parts of the section gave the amount of starch



present. By removing the twig to a bath of alcohol for a minute the subject was cleared up, and the location of the starch-bearing portions could be made out more distinctly. If desired, thin sections could be taken from the treated surface for microscopic study.

**SUGARS.**—The sugars of various sorts are usually associated with each other, and only the presence or absence of the group was usually attempted. Trommer's test is a simple and satisfactory one: Place the slices or sections in a porcelain dish, add a strong solution of cupric sulphate and heat to boiling. Wash the specimens thoroughly, and add hot potassic hydrate, when the presence of dextrine and grape-sugar will be shown by a reddish precipitate. To separate these two sugars, treat the original tissue to alcohol, standard strength, for a few hours, which will dissolve away the grape-sugar, when the dextrine can be tested for as before. Cane-sugar, with Trommer's, gives a bright blue color in the cell contents, quickly vanishing, but no red precipitate. It is well for the student to make tests with this reagent of vegetable tissues containing large quantities of each one of the leading kinds of sugars. Fehling's solution, which is a modification of the above, may be employed. In place of the pure cupric sulphate solution, one of one part of the cupric sulphate and five parts of potassium sodium tartrate in eight parts of water may be used. This reagent needs to be kept in the dark.

**ALBUMINOIDS.**—For these Millon's reagent was employed. This is prepared by adding to metallic mercury an equal weight of concentrated nitric acid. When all is dissolved add twice the volume of pure water. Place the tissue in a porcelain dish, add a little of the reagent and heat, when a rose or brick-red precipitate indicates the presence of albuminoids. A dark yellow, with nitric acid, and a yellow or brown with iodine also indicate the same substances.

**CRYSTALS.**—There are two leading salts in the form of crystals in plants, the carbonate of lime and the oxalate. The crystals having been detected with the microscope, acetic acid may be added, which will dissolve the carbonate with effervescence, but does not act upon the oxalate.

**TANNIN.**—The various iron salts give with tannin a very



dark color, therefore ferric chloride may be employed for its detection.

LIGNIN.—Carbolic acid dissolved in concentrated hydrochloric acid gives a green color with lignin, and hydrochloric acid and alcohol develop a beautiful rose color after some hours.

The works most frequently consulted for methods in the studies contained in this paper have been "Behren's Guide to the Microscope in Botany," by Hervey, and "Goodale's Physiological Botany." Upon the general subject much assistance can be obtained from "Sach's Vegetable Physiology," "Vine's Physiology of Plants," and "Strasburger's Das Botanische Practicum." The most elaborate paper consulted is a "Memoire sur la Moelle des Plantes Ligneuses," with numerous elegant plates, by A. Gris, in "Nouvelle Archives du Museum d'Histoire Naturelle, 1870."

RUTGERS COLLEGE,

July 21st, 1890.